

SDS-PAGE Analysis of *Mycobacterium Avium* Paratuberculosis Whole Cell Extracts

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Background & Objectives: *Mycobacterium avium* subsp. paratuberculosis (MAP) cause granulomatous enteritis of ruminants with economical losses to the dairy industry. Ruminants affected by JD develop strong immunological reactivity against *M. avium* subsp. paratuberculosis antigens. A whole range of new antigens has been identified and characterized. Some of these antigens are now being evaluated for the development of new diagnostics for paratuberculosis. A36 is the major complex of *Mycobacterium avium* paratuberculosis. The complex contains some 30 proteins (20 to 90 kDa), whose immunological activities in humoral and cellular immunity have been explored. In this study we used the whole cell extract of MAP to evaluate the molecular weight of proteins.

Methods: The soluble protein extracts were separated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) under reducing conditions as described by Laemmli. Electrophoresis was carried out at a constant current of 120 V for 75 min. The protein bands were stained by Coomassie brilliant blue staining solution. Each protein was quantified for molecular net intensity and the molecular weights were established on the bases of mobility compared to that of standard molecular weight marker (Bio-Rad).

Results: The SDS-PAGE of MAP extracts shows 6 bands with molecular weight 65, 53, 42, 34, 25, 17.

Conclusion: Several antigenic proteins have been described in the pathogen identified. Components in the 28-kDa to 45-kDa range are the most antigenic for both cellular immunity and humoral immunity. So identification and characterization of these components may be useful for understanding the pathogenic mechanisms of MAP and the immune response against this organism. In this study SDS-PAGE of MAP extracts showed 6 bands that these bands could be antigenic for host immunity and we can isolate these proteins for more study.

Keywords: SDS-PAGE; *Mycobacterium avium paratuberculosis*; Whole Cell Extracts